Repeated implantation failure: clinical approach

Alex Simon, M.D., and Neri Laufer, M.D.

In Vitro Fertilization Unit, Department of Obstetrics and Gynecology, Ein Kerem, Hebrew University, Hadassah Medical Center, Jerusalem, Israel

Successful embryo implantation depends on a well-functioning endometrium as well as a normal healthy embryo. This process might be hampered if either of these variables is defective. Repeated implantation failure (RIF) is diagnosed when good-quality embryos repeatedly fail to implant after transfer in several IVF treatment cycles. The causes of RIF originate with either the mother or the embryo. The authors discuss factors that are associated with RIF and address various treatment options. (Fertil Steril® 2012;97:1039–43. ©2012 by American Society for Reproductive Medicine.)
Key Words: Repeated implantation failure, recurrent implantation failure, RIF, IVF, implantation

Repeated implantation failure (RIF) is determined when transferred embryos fail to implant after several IVF treatment attempts. However, there are no formal criteria defining the number of failed cycles or the total number of embryos transferred in these IVF attempts. Accordingly, different centers practicing IVF may use different definitions for RIF (1). Considering the current success rate of IVF treatment and the mean number of embryos transferred in each cycle, we recommend defining RIF as failure of implantation in at least three consecutive IVF attempts, in which one to two embryos of high-grade quality are transferred in each cycle.

Successful implantation is a complex process involving two main players: the mother as a host and the embryo. Problems originating from the host environment, such as abnormal uterine anatomy, nonreceptive endometrium, and the medical condition of the mother (such as thrombophilia and abnormal immunologic response) can adversely affect the cross-talk between the embryo and the endometrium that is so crucial for successful implantation (2–5). Similarly, this endometrial–embryo interaction may be hampered if the embryo is disordered. Embryo abnormality can originate from either paternal sperm factors or from the oocyte and its ability to be fertilized normally and cleave. Accordingly, the investigation and treatment of patients with RIF should focus on both male and female risk factors that, once identified, should be managed and treated appropriately (Fig. 1).

FEMALE FACTORS AND RIF

Anatomic Causes

After several consecutive IVF failures and in agreement with the definition of RIF, patients should undergo hysteroscopy to assess the uterine cavity. Three-dimensional ultrasonography and hysterosalpingography are complementary tools to be performed as needed. Once an abnormality associated with implantation failure is recognized, treatment options should be considered, including uterine septectomy, removal of intrauterine adhesions, endometrial polypectomy or myomectomy, particularly the submucous type, and excision of hydrosalpinx (5–7).

Endometrium

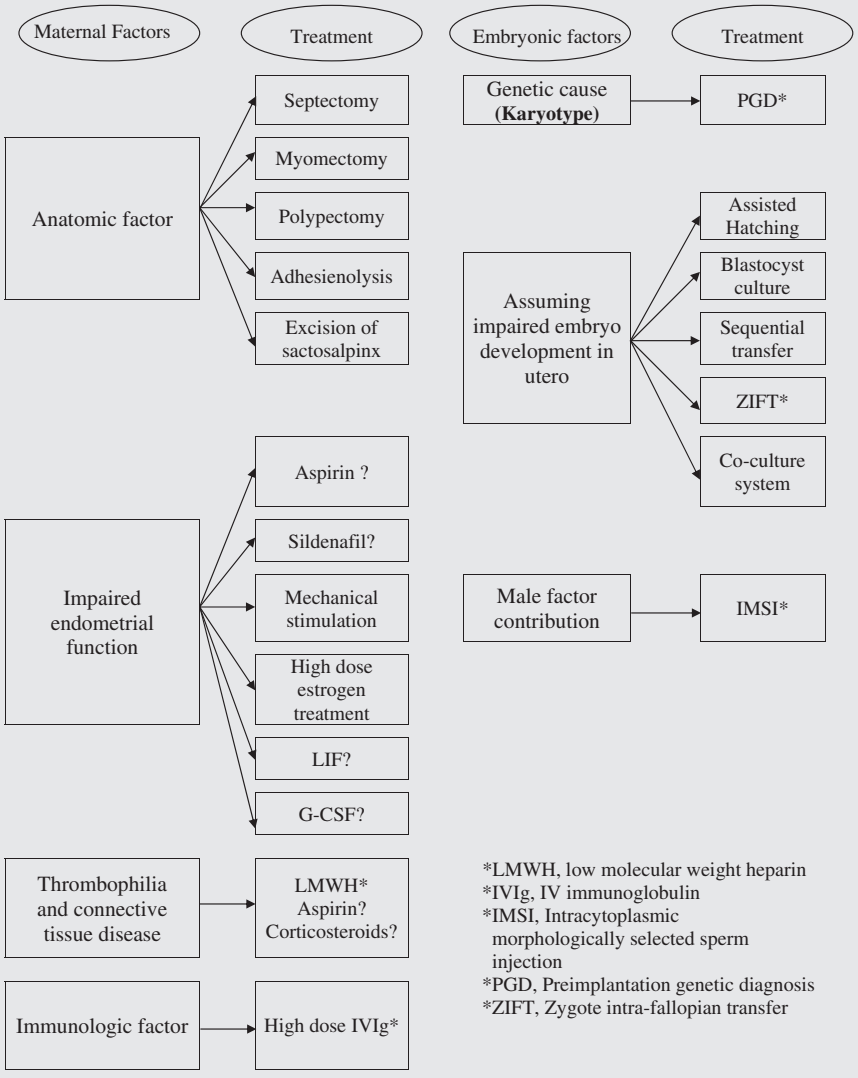
A functioning and receptive endometrium is crucial for embryo implantation. During the menstrual cycle, the endometrium undergoes both morphologic and biologic changes, during which it becomes prepared for interaction with the embryo, leading to successful implantation. Once all biological changes are adequate, the embryo can attach, invade the endometrium, and finally implant. Ultrasound examination of the thickness and appearance of the endometrium is an easily performed means of assessing morphologic changes occurring in the endometrium during the follicular phase, and is thus used as a measure to predict successful implantation. Indeed, several studies have reported a strong association between endometrial thickness and successful implantation (8, 9). Noyes et al. (9) reported a significantly higher pregnancy rate of 48.6% in patients with endometrial thickness of >9 mm, as compared with 16% in those with <9 mm. Nevertheless, others failed to confirm such an association (10, 11). The minimal adequate endometrial thickness for successful implantation, as measured in the late proliferative phase, varies between studies, with a range of 6–8 mm. Thin, unresponsive endometrium is hard to manage (12), and if all

Received February 1, 2012; revised March 8, 2012; accepted March 9, 2012; published online March 30, 2012.
A.S. has nothing to disclose. N.L. has nothing to disclose.
Reprint requests: Alex Simon, M.D., Hadassah Medical Center, Department of Obstetrics and Gynecology, Ein Kerem, P.O. Box 12000, Jerusalem 91120, Israel (E-mail: simonal@hadassah.org.il).

Fertility and Sterility® Vol. 97, No. 5, May 2012 0015-0282/$36.00

doi:10.1016/j.fertnstert.2012.03.010

FIGURE 1



Evaluation and treatment of RIF.

Simon. RIF: clinical approach. Fertil Steril 2012.

available treatments (i.e., high dose of estrogen, the application of vaginal estrogen pills, aspirin, and other medications that may increase blood flow to the endometrium) fail, then surrogacy is a reasonable option (1).

Several investigators have suggested that patients with RIF may benefit from mechanical endometrial stimulation performed in the cycle preceding the actual treatment cycle. This local injury is induced by means of an endometrial biopsy catheter and sampling (13, 14). It is postulated that the biopsy induces an inflammatory response that may facilitate the preparation of the endometrium for implantation (15). The exact mechanism of such a positive effect on implantation after endometrial sampling, as well as the number and timing of endometrial stimulations in the cycle preceding that of ET, needs to be determined.

To facilitate implantation, and on the basis of information elaborating the role of some mediators participating in this process, several investigators systemically administered human leukemia inhibitory factor. However, such treatment did not improve implantation or pregnancy outcomes (16). Others administered granulocyte colony-stimulating factor, either locally (17) or systemically (18), and found the treatment useful in improving IVF outcome in patients with unexplained RIF. Although encouraging, more information is needed before suggesting this treatment modality on a routine basis to patients with RIF.

Thrombophilia

In recurrent pregnancy loss, patients are advised to undergo blood tests for thrombophilia, as well as for connective tissue diseases that involve antiphospholipid antibodies. However, thrombophilia and antiphospholipids may also be associated with risk for RIF (19, 20). Once detected, a consultation with

a hematologist and connective tissue disease specialist is advocated, and treatment with low molecular weight heparin (LMWH) is recommended. When a thrombophilic trait is detected, treatment with a prophylactic dose of LMWH is sufficient and seems to improve IVF outcome (21, 22). However, when antiphospholipid antibody syndrome is diagnosed, a concomitant treatment with a mini-dose of aspirin and/or corticosteroids should be considered. Once a hypercoagulability state is diagnosed, the appropriate treatment protocol for ovarian stimulation should be implemented to minimize the risk of ovarian hyperstimulation syndrome. Initiation of LMWH should be considered from the early stimulation period or from the day of ET. The approach should be determined after consultation. A patient's family and personal medical history, particularly her previous IVF experience, are important for reaching a decision. Patients with no history of thrombotic events, personally or among close relatives, and who already experienced several uneventful IVF treatments, may be considered suitable to start LMWH on the day of ET. Patients with antiphospholipid antibody syndrome, or with a history of a disease that can be attributed to a hypercoagulability trait, should start anticoagulation concomitant with gonadotropin administration. Treatment with LMWH should be stopped 24 hours before egg retrieval and reinitiated the day after ovum pick-up. Empirical treatment with LMWH, aspirin, or corticosteroids was not found to be effective, and is not advocated for women with RIF who were negative for thrombophilic tests (23, 24).

EMBRYONIC FACTORS

Even embryos that are morphologically defined to be of good quality may cease to develop in utero and fail to progress to the blastocyst stage, resulting in failure of implantation. This may be due to either suboptimal local conditions or intrinsic factors within the embryos. Among the approaches that have demonstrated effectiveness in overcoming these obstacles are zygote intrafallopian transfer (25, 26), blastocyst transfer (27, 28), sequential embryo transfer (29, 30), and embryo coculture (31, 32).

Hatching of the blastocyst through the zona pellucida is an essential step preceding implantation. Assisted hatching was suggested to increase implantation rates in patients who had previously failed IVF cycles. After reviewing the literature, the American Society for Reproductive Medicine Practice Committee concluded that the currently available published evidence does not support the routine or universal application of assisted hatching in all IVF cycles (33). According to the guidelines discussed by the committee, we use laser-assisted hatching in patients with RIF, in those with poor embryo quality, and in women of an advanced age (>38 years).

Genetics

A couple diagnosed with RIF should have karyotype testing to rule out structural anomalies of chromosomes. Although structural disorders generally lead to habitual abortions, they may also prohibit implantation (34). If a structural anomaly is detected, then preimplantation genetic diagnosis should be offered. Comparative genomic hybridization (CGH) testing should not be advocated for the couple, because it cannot detect balanced translocations.

Similarly, the efficacy of preimplantation genetic screening of early cleaved embryos in couples with normal karyotypes has not been substantiated and therefore, at present, this technique should not be applied for RIF patients (35). In the future, when higher rates of blastocysts will be obtainable in culture media and the new freezing method of vitrification will be routinely practiced, array CGH applied on a blastocyst-stage biopsy to detect minor differences in DNA content should be considered for RIF, to better choose the embryo for transfer (35).

Sperm Contribution

Investigation of causes of RIF may include some forms of advanced morphologic analysis of the sperm, because the contribution of the sperm cell to the production of normal and healthy embryos is crucial. Testing sperm cells for DNA fragmentation and abnormal chromatin packaging is reasonable if RIF seems to be associated with male factor. This test, however, should also be carried out in patients with apparently normal sperm parameters. If advanced morphologic evaluation of the sperm or other methods of assessing DNA integrity reveal a high percentage of abnormal sperm cells, intracytoplasmic morphologically selected sperm injection (IMSI) should be considered as a means of improving implantation (36, 37). In a recent prospective randomized study, Balaban et al. (38) reported higher implantation and clinical pregnancy rates for patients who had IMSI as compared with those who had intracytoplasmic sperm injection, 28.9% vs. 19.5% and 54.0% vs. 44.4%, respectively. We routinely use the IMSI procedure in severe cases of RIF, defined as five or more IVF failures.

Immunologic Factor

If the results of all tests mentioned above are normal, consideration of a possible contribution of the couple's immunologic system to implantation failure is recommended. This can first be performed by checking for the presence of an immunologic reaction after a mutual cross-match between the serum and lymphocytes of the couple. If no reaction results, then the maternal immune system is apparently nonresponsive to paternal antigen components. This may be due to the couple's similarity in human leukocyte antigen (HLA) components. In such case, a similarity of alleles in class I and class II HLA compatibility should be tested. Human leukocyte antigen similarity is reported to be associated mainly with recurrent abortions. However, in the extreme situation, it may interfere with implantation, because dissimilarity of HLA is also crucial at the very beginning of the implantation process, during which the immune system plays an important role. If such a similarity is found, high-dose IV immunoglobulin should be offered before ET, as suggested by us (39), followed by an additional dose as soon as a heartbeat is visualized, at approximately 6 weeks' gestation.

FUTURE CONSIDERATIONS

Better selection of the embryo for transfer is expected once new methods of time-lapse imaging (40) and "omics"

technology (41, 42) are applied. These methods can accurately assess embryo morphology and its metabolic activity. In the future, within the implementation of these modalities, implantation rates for IVF patients, including those with RIF, are expected to increase. "Omic" technologies may also improve our understanding of the processes involved in the endometrium during implantation (43, 44). This will promote better recognition of the "window of implantation" and may suggest options as to how manipulate this crucial period, to facilitate the cross-talk between the embryo and its platform, resulting in improved implantation rates.

Better culture conditions and increased success rates of embryo freezing by vitrification, as well as innovative methods in molecular genetic biology such as array CGH, will help achieve the goal of obtaining normal, healthy embryos, capable of implanting after transfer.

REFERENCES

1. Shufaro Y, Schenker JG. Implantation failure, etiology, diagnosis and treatment. Int J Infertil Fetal Med 2011;2:1–7.
2. Fukui A, Funamizu A, Yokota M, Yamada K, Nakamua R, Fukuhara R, et al. Uterine and circulating natural killer cells and their roles in women with recurrent pregnancy loss, implantation failure and preeclampsia. J Reprod Immunol 2011;90:105–10.
3. Achache H, Revel A. Endometrial receptivity markers, the journey to successful embryo implantation. Hum Reprod Update 2006;12:731–46.
4. Gonen-Gross T, Goldman-Wohl D, Huppertz B, Lankry D, Greenfield C, Natanson-Yaron S, et al. Inhibitory NK receptor recognition of HLA-G: regulation by contact residues and by cell specific expression at the fetal-maternal interface. PLoS One 2010;5:e8941.
5. Margalioth EJ, Ben-Chetrit A, Gal M, Eldar-Geva T. Investigation and treatment of repeated implantation failure following IVF-ET. Hum Reprod 2006; 21:3036–43.
6. Practice Committee of American Society for Reproductive Medicine, Society of Reproductive Surgeons. Salpingectomy for hydrosalpinx prior to in vitro fertilization. Fertil Steril 2008;90(5 Suppl):S66–8.
7. Demirol A, Gurgan T. Effect of treatment of intrauterine pathologies with office hysteroscopy in patients with recurrent IVF failure. Reprod Biomed Online 2004;8:590–4.
8. Dickey RP, Olar TT, Curole DN, Taylor SN, Rye PH. Endometrial pattern and thickness associated with pregnancy outcome after assisted reproduction technologies. Hum Reprod 1992;7:418–21.
9. Noyes N, Liu HC, Sultan K, Schattman G, Rosenwaks Z. Endometrial thickness appears to be a significant factor in embryo implantation in in vitro fertilization. Hum Reprod 1995;10:919–22.
10. Khalifa E, Brzyski RG, Oehninger S, Acosta AA, Muasher SJ. Sonographic appearance of the endometrium: the predictive value for the outcome of in vitro fertilization in stimulated cycles. Hum Reprod 1992;7:677–80.
11. Oliveira JB, Baruffi RL, Mauri AL, Petersen CG, Campos MS, Franco JG Jr. Endometrial ultrasonography as a predictor of pregnancy in an in vitro fertilization programme. Hum Reprod 1993;8:1312–5.
12. Shufaro Y, Simon A, Laufer N, Fatum M. Thin unresponsive endometrium: a possible complication of surgical curettage compromising ART outcome. J Assist Reprod Genet 2008;25:421–5.
13. Raziel A, Schachter M, Strassburger D, Bern O, Ron-El R, Friedler S. Favorable influence of local injury to the endometrium in intracytoplasmic sperm injection patients with high order implantation failure. Fertil Steril 2007;87: 198–201.
14. Narvekar SA, Gupta N, Shetty N, Kottur A, Srinivas M, Rao KA. Does local endometrial injury in the nontransfer cycle improve the IVF-ET outcome in the subsequent cycle in patients with previous unsuccessful IVF? A randomized controlled pilot study. J Hum Reprod Sci 2010;3: 15–9.
15. Gnainsky Y, Granot I, Aldo PB, Barash A, Or Y, Schechtman E, et al. Local injury of the endometrium induces an inflammatory response that promotes successful implantation. Fertil Steril 2010;94:2030–6.
16. Brinsden PR, Alam V, de Moustier B, Engrand P. Recombinant human leukemia inhibitory factor does not improve implantation and pregnancy outcomes after assisted reproductive techniques in women with recurrent unexplained implantation failure. Fertil Steril 2009;91(4 Suppl):1445–7.
17. Gleicher N, Vidali A, Barad DH. Successful treatment of unresponsive thin endometrium. Fertil Steril 2011;95:2123.e13–7.
18. Würfel W, Santjohanser C, Hirv K, Bühl M, Meri O, Laubert I, et al. High pregnancy rates with administration of granulocyte colony-stimulating factor in ART-patients with repetitive implantation failure and lacking killer-cell immunoglobulin-like receptors. Hum Reprod 2010;25:2151–2.
19. Azem F, Many A, Ben Ami I, Yovel I, Amit A, Lessing JB, et al. Increased rates of thrombophilia in women with repeated IVF failures. Hum Reprod 2004; 19:368–70.
20. Grandone E, Colaizzo D, Lo Bue A, Checola MG, Cittadini E, Margaglione M. Inherited thrombophilia and in vitro fertilization implantation failure. Fertil Steril 2001;76:201–2.
21. Bohlmann MK. Effects and effectiveness of heparin in assisted reproduction. J Reprod Immunol 2011;90:82–90.
22. Qublan H, Amarin Z, Dabbas M, Farraj AE, Beni-Merei Z, Al-Akash H, et al. Low-molecular-weight heparin in the treatment of recurrent IVF-ET failure and thrombophilia: a prospective randomized placebo-controlled trial. Hum Fertil (Camb) 2008;11:246–53.
23. Seshadri S, Sunkara SK. Low-molecular-weight-heparin in recurrent implantation failure. Fertil Steril 2011;95:e29.
24. Berker B, Taşkin S, Kahraman K, Taşkin EA, Atabekoğlu C, Sönmezer M. The role of low-molecular-weight heparin in recurrent implantation failure: a prospective, quasi-randomized, controlled study. Fertil Steril 2011;95: 2499–502.
25. Levran D, Mashiach S, Dor J, Levron J, Farhi J. Zygote intrafallopian transfer may improve pregnancy rate in patients with repeated failure of implantation. Fertil Steril 1998;69:26–30.
26. Farhi J, Weisman A, Nahum H, Levran D. Zygote intrafallopian transfer in patients with tubal factor infertility after repeated failure of implantation with in vitro fertilization-embryo transfer. Fertil Steril 2000;74:390–3.
27. Guerif F, Bidault R, Gasnier O, Couet ML, Gervereau O, Lansac J, et al. Efficacy of blastocyst transfer after implantation failure. Reprod Biomed Online 2004;9:630–6.
28. Levitas E, Lunenfeld E, Har-Vardi I, Albotiano S, Sonin Y, Hackmon-Ram R, et al. Blastocyst stage embryo transfer in patients who failed to conceive in three or more day 2–3 embryo transfer cycles: a prospective randomized study. Fertil Steril 2004;81:567–71.
29. Loutradis D, Drakakis P, Dallianidis K, Bletsa SR, Milingos S, Doumplis N, et al. A double embryo transfer on days 2 and 4 or 5 improves pregnancy outcome in patients with good embryos but repeated failures in IVF or ICSI. Clin Exp Obstet Gynecol 2004;31:63–6.
30. Almog B, Levin I, Wagman I, Kapustiansky R, Schwartz T, Mey-Raz N, et al. Interval double transfer improves treatment success in patients with repeated IVF/ET failures. J Assist Reprod Genet 2008;25:353–7.
31. Spandorfer SD, Pascal P, Parks J, Clark R, Veeck L, Davis OK, et al. Autologous endometrial coculture in patients with IVF failure: outcome of the first 1,030 cases. J Reprod Med 2004;49:463–7.
32. Eyheremendy V, Raffo FG, Papayannis M, Barnes J, Granados C, Blaquier J. Beneficial effect of autologous endometrial cell coculture in patients with repeated implantation failure. Fertil Steril 2010;93:769–73.
33. Practice Committee of Society for Assisted Reproductive Technology, Practice Committee of American Society for Reproductive Medicine. The role of assisted hatching in in vitro fertilization: a review of the literature. A Committee opinion. Fertil Steril 2008;90(5 Suppl):S196–8.
34. Raziel A, Friedler S, Schachter M, Kasterstein E, Strassburger D, Ron-El R. Increased frequency of female partner chromosomal abnormalities in patients with high-order implantation failure after in vitro fertilization. Fertil Steril 2002;78:515–9.
35. Harper JC, Sengupta SB. Preimplantation genetic diagnosis: state of the ART 2011. Hum Genet 2012;131:175–86.

36. Souza Setti A, Ferreira RC, Paes de Almeida Ferreira Braga D, de Cássia Sávio Figueira R, Iaconelli A Jr, Borges E Jr. Intracytoplasmic sperm injection outcome versus intracytoplasmic morphologically selected sperm injection outcome: a meta-analysis. Reprod Biomed Online 2010;21:450–5.
37. Wilding M, Coppola G, di Matteo L, Palagiano A, Fusco E, Dale B. Intracytoplasmic injection of morphologically selected spermatozoa (IMSI) improves outcome after assisted reproduction by deselecting physiologically poor quality spermatozoa. J Assist Reprod Genet 2011;28:253–62.
38. Balaban B, Yakin K, Alatas C, Oktem O, Isiklar A, Urman B. Clinical outcome of intracytoplasmic injection of spermatozoa morphologically selected under high magnification: a prospective randomized study. Reprod Biomed Online 2011;22:472–6.
39. Elram T, Simon A, Israel S, Revel A, Shveiky D, Laufer N. Treatment of recurrent IVF failure and human leukocyte antigen similarity by intravenous immunoglobulin. Reprod Biomed Online 2005;11:745–9.
40. Meseguer M, Herrero J, Tejera A, Hilligsøe KM, Ramsing NB, Remohí J. The use of morphokinetics as a predictor of embryo implantation. Hum Reprod 2011;26:2658–71.
41. Assou S, Boumela I, Haouzi D, Anahory T, Dechaud H, De Vos J, et al. Dynamic changes in gene expression during human early embryo development: from fundamental aspects to clinical applications. Hum Reprod Update 2011;17:272–90.
42. Hardarson T, Ahlström A, Rogberg L, Botros L, Hillensjö T, Westlander G, et al. Non-invasive metabolomic profiling of day 2 and 5 embryo culture medium: a prospective randomized trial. Hum Reprod 2012;27:89–96.
43. Haouzi D, Dechaud H, Assou S, De Vos J, Hamamah S. Insights into human endometrial receptivity from transcriptomic and proteomic data. Reprod Biomed Online 2012;24:23–4.
44. Díaz-Gimeno P, Horcajadas JA, Martínez-Conejero JA, Esteban FJ, Alamá P, Pellicer A, et al. A genomic diagnostic tool for human endometrial receptivity based on the transcriptomic signature. Fertil Steril 2011;95:50–60, 60.e1–15.